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OBSERVATIONS ON HETEROSTYLOUS PLANTS

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(WITH PLATES XXI–XXIII)

The discovery of the "sex determinant" or "accessory chromosome" in the sperms of certain insects is probably the most notable cytological advance of the present decade. Furnishing, as it apparently does, a cytological basis for the predestination of sex at the time of fertilization, it has an important bearing on the whole question of the determination and heredity of sex. The condition found in these insects has been too widely discussed to need description here. It may be briefly summed up, however, as follows: examination has shown that the sperms are of two classes, equal in number, which differ in respect to one or more of the chromosomes which enter into the formation of their nuclei; and the facts clearly demonstrate that fertilization of the eggs by one class produces males, by the other class females. This difference, the significance of which was first suggested by MCCLUNG in 1902, has been shown to occur in nearly a hundred species of insects (WILSON **40,** p. 57).

Its prevalence here has suggested the possibility of a similar condition in all animals having separate sexes. Attempts have also been made to demonstrate such a condition in dioecious plants, but as yet no positive results have been obtained. Miss SYKES published in 1909 (**39**) a brief note on the nuclei of some dioecious plants. She studied *Hydrocharis Morsus-ranae, Bryonia dioica, Lychnis dioica, Mercurialis perennis,* and *Sagittaria montevidensis,*

and states that the nuclei of the male and female plants were in all cases apparently identical, both in the number and in the character of their chromosomes. More complete studies have been made by DARLING (5) in *Acer Negundo*, and by STRASBURGER (38) in *Melandrium rubrum, Cannabis sativa, Mercurialis dioica*, and *Bryonia dioica*. Both writers report that in the reduction division of the microspore mother cells they find no evidence of anything which might be considered a "sex determinant." Negative results in these few cases, of course, do not prove that a sex determinant never occurs in plants, for it will be remembered that such a condition has been demonstrated in only a comparatively few animals.

Dimorphic heterostylous plants present, as will be pointed out below, such striking resemblances to dioecious plants that the present study was undertaken in the hope of finding, in the reduction division of the microspore mother cells, some indication of a morphological difference in the chromosomes which go to make up the nuclei of the microspores.

Material of several species was prepared, but only two, *Fagopyrum esculentum* and *Houstonia caerulea*, proved favorable for study. The material used was all collected in the spring and summer of 1910. The buds of *Fagopyrum* were taken from vigorous plants under cultivation; those of *Houstonia* were nearly all from an old, well-established lawn, where many of the clumps had very likely persisted for several years.

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Historical sketch

PERSOON, in 1794, notes that in certain species of *Primula* there are two forms which exist in about equal numbers and differ from each other in the length of their pistils and stamens. This is, according to VON MOHL (p. 326), the first description of the condition now known as heterostyly.

This condition was apparently regarded as a mere variation in form until the time of DARWIN, who discusses several dimorphic

and trimorphic genera in a series of papers read before the Linnean Society during the years 1862–1868. In these early papers Darwin (**6–10**) calls attention to the fact that the morphological differences in the flowers constitute a device by which cross pollination is favored, and that these differences in form are associated with physiological differences which affect their fertility, so that a flower of either form is more likely to be fully fertilized when pollinated from a flower of the other form ("legitimate pollination") than when pollinated from a flower of the same form ("illegitimate pollination"). He also describes the offspring of illegitimate unions and points out that they differ from normal plants and have what he calls a "hybrid-like" nature.

Darwin afterward collected these papers and published them in a "connected and corrected form, together with new matter" in *The different forms of flowers in plants of the same species*, which contains also an account of the literature on the subject up to the date of its publication (1877). Only three of the writers whom he mentions, Hildebrand, Scott, and Müller, treat heterostyly as anything more than a mere difference in form or at most a device to favor cross pollination.

Hildebrand, who first used the term "heterostyled," was also the first to investigate the inheritance of heterostyly. In his first paper (**18**) he describes experiments on the illegitimate fertilization of *Linum perenne* and *Primula sinensis*. The illegitimately pollinated flowers of *Linum* were uniformly sterile. In *Primula*, however, all the illegitimately pollinated flowers developed capsules, which contained an average of 18 seeds, about two-fifths the number found in the capsules of legitimately pollinated flowers. Hildebrand planted the seeds thus produced, and found that while the seeds of either form legitimately fertilized produce long-styled and short-styled forms in about equal numbers, the seeds from illegitimate unions tend to reproduce the parent form. Seeds from illegitimate unions of long-styled plants, however, tend to transmit the parent form more truly than those of the short-styled. This conclusion was accepted by Darwin, but the results he later obtained from similar experiments with *Primula sinensis* do not agree very closely with those of Hildebrand.

Two later papers by HILDEBRAND (**19, 20**) deal with trimorphous species of *Oxalis*. He found in *Oxalis Valdiviana* (**20,** p. 43) that seeds from any one of the six possible legitimate unions produced all three forms, but that the two parent forms greatly predominated. He also states that long-styled plants of *Oxalis rosea*, growing by themselves, have always produced long-styled plants ("Jahr aus Jahr ein dieselbe Form entsteht").

In 1864, JOHN SCOTT published a paper (**32**) in which he arranges all the known species of *Primula* in four groups: dimorphic, short-styled, long-styled, and non-dimorphic (homostyled), and describes experiments with 7 dimorphic species. He pollinated each of these 7 species both legitimately and illegitimately and counted the seeds obtained by each method. The result was uniformly that legitimate unions produced a markedly greater number of seeds than illegitimate.

FRITZ MÜLLER (**27**), in a brief paper dealing with a trimorphous species of *Pontederia* growing in Brazil, mentions the fact that in *Oxalis Regnelli*, another trimorphous species, the seeds of long-styled plants, legitimately fertilized with pollen from the longest stamens of the mid-styled form, produced plants which belonged exclusively to the two parent forms.

In his *Forms of flowers*, DARWIN includes all that had been done on heterostylous plants up to the time of its publication. He cites 38 genera known to include heterostyled species. These genera are distributed as follows (**11,** p. 254): Hypericineae 1, Erythroxyleae 2, Geraniaceae 2, Lythraceae 2, Rubiaceae 17, Primulaceae 3, Oleaceae 1, Gentianaceae 3, Polemoniaceae 1, Cordieae 1, Boragineae 1, Verbenaceae 1, Polygoneae 1, Thymeleae 1, and Pontederiaceae 1. The wide geographical distribution of the genera which contain heterostyled species and the fact that the families to which they belong are mostly very distinct from one another, indicate that heterostyly has arisen independently in several phylogenetic lines.

DARWIN considers (p. 245) that the morphological differences between the forms of a heterostyled species are confined to the flower. His observations on this point may be summed up briefly as follows: In the calyx there are no differences. The corolla

shows slight differences in shape due to the different position of the anthers.   In *Pulmonaria* there is also a slight difference in the size of the corolla, and in *Pontederia* in its color.   The most striking differences, of course, occur in the stamens and pistils (p. 247). The pistils differ in length of style and in size and shape of stigma. The stamens show a corresponding difference in length of filament or in place of insertion; and there is sometimes a difference in the color and thickness of the filaments, and in the size of the anthers. The pollen grains usually show a marked difference in size in the different forms.   Of 43 cases cited, only 8 showed pollen grains of equal size in the different forms.   In all the others the size of the pollen in the short-styled form exceeded that of the long-styled form (p. 249).   The greatest difference was found in a tri-morphous species of *Pontederia*, in which the diameters of the pollen grains from the longest stamens are to those of the shortest as 100:55, indicating a difference in contents in the ratio of 6:1.

These morphological differences are slight, however, compared with the physiological differences which accompany them.   Repeated experiments have shown that complete fertility in heterostylous plants is secured only when a flower is pollinated with pollen from a flower of another form, that is, the pollen must come from a stamen equal in length to the pistil on which it is placed.   The superiority of legitimate over illegitimate pollination is shown by the propor-tion of flowers which yield capsules and by the average number of seeds per capsule (p. 245).   As DARWIN himself states, mor-phological characters alone do not furnish conclusive evidence of heterostyly.   Final proof can be derived only from experiments which show that pollen must be applied from one form to the other in order to insure complete fertility.

The physiological difference in the forms is exhibited also in the time necessary to secure fertilization with legitimate and illegiti-mate pollen.   DARWIN (p. 31) placed on several stigmas of a long-styled cowslip plenty of pollen from the same plant, and after 24 hours added some from a short-styled dark red polyanthus.   From the flowers thus treated, 30 seedlings were raised, and all, without exception, bore reddish flowers.   DARWIN describes a still more striking difference in *Linum perenne* (p. 87).   He placed pollen

from a long-styled flower on all 5 stigmas of a long-styled flower on a separate plant. After 19 hours the stigmas were dissected and only a single pollen grain had emitted a tube. The pollen proved to be good when placed on the stigma of a short-styled plant. This experiment was repeated three times, with uniform results. A similar condition has been shown to occur in *Lythrum Salicaria* by STRASBURGER (**34,** p. 82). In this plant illegitimate pollination resulted in only a very slight growth of the pollen tube.

Physiological differences appear also in ways which less directly affect fertilization. In the long-styled form of *Linum perenne* (DARWIN, **11,** p. 130), each separate stigma rotates on its own axis when the flower is mature, thus turning its papillose surface outward. This movement is confined to the long-styled form. In *Faramea* the stamens of the short-styled form rotate on their axes. No such motion is found in the stamens of the long-styled form.

DARWIN's experiments on the inheritance· of heterostyly, like those of the other earlier workers, do not give very uniform results. But he deduces the general laws that seeds from illegitimate unions tend to reproduce the parent form (p. 271), and that illegitimate unions of long-styled plants tend to transmit the parent form more truly than do those of short-styled plants.

Recently, BATESON and GREGORY (**2**) have experimented on the inheritance of heterostyly in *Primula*. They find that in *Primula sinensis* the inheritance follows the Mendelian type, the short style being the dominant character and the long style the recessive. Short-styled plants are then heterozygotes, and half their gametes bear the dominant character, the other half the recessive; while long-styled plants are homozygotes and all their gametes bear the recessive character. One remarkable exception, however, was found. This was a single short-styled plant in which the female gametes were normal, that is, half bore the dominant and half the recessive character, while the male gametes bore the dominant character almost exclusively. BATESON and GREGORY note, as did DARWIN, that about half the eggs are fertilized by illegitimate pollen, while the rest are not; and suggest that this may be due to a differentiation of the egg cells of the plants.

ERRERA (**13**) has pointed out that *Primula elatior* shows what

he calls "caractères hétérostyliques secondaires." That is, the two forms differ not only in the parts of the flower, but also in the forms of the leaves. As he describes them (p. 229), the leaves of the long-styled form are "relatively longer and narrower, the ratio of the mean length (measured from the base of the petiole to the tip of the blade) to the maximum width being 2.86:1, and in plants grown in deep shade, 3.63:1"; while the leaves of the short-styled form are "relatively wider and shorter, the ratio of the mean length to the maximum width being 2.41:1, and in plants grown in the shade, 3.11:1."

Relation of heterostyly to dioeciousness

Dimorphic heterostylous plants present, in several respects, a striking resemblance to dioecious forms. In other hermaphrodite plants and in hermaphrodite animals, there is presumably unlimited possibility of crossing. In dimorphic heterostyled plants, however, the individuals are divided into two classes, which exist in approximately equal numbers and are adapted for reciprocal fertilization, a condition essentially the same as that found in dioecious plants and in the higher animals. This resemblance is made still more evident by ERRERA's recent discovery of differences in the vegetative organs of the two forms in *Primula elatior*, comparable to the secondary sexual characters common in animals and found in a few dioecious plants, such as the hemp.

Naturally, no very definite comparison can be drawn between the inheritance of heterostyly and the inheritance of sex until it is decided what laws the inheritance of sex actually follows. But it may at least be pointed out that the condition described by BATESON and GREGORY for *Primula sinensis*, in which one form is a heterozygous dominant and the other a homozygous recessive, is exactly the condition believed, by several workers, to exist in the inheritance of sex, notably by CORRENS (4) for the dioecious *Bryonia alba*, and by BATESON for animals (Wilson 40, p. 63). Aside from any analogy with dioecious plants, the work of BATESON and GREGORY on *Primula sinensis* indicates that in this form, at least, the inheritance follows the Mendelian law, a condition which indicates that a segregation of different characters occurs in the

reduction division of both megaspores and microspores of one form, in this case the short-styled form. Whether this is accompanied by any morphological difference in the chromosomes or not is of course another question.

The resemblance of dimorphic heterostylous plants to dioecious plants suggested to DARWIN (p. 285) that heterostyly may have been one of the ways by which the dioecious condition among flowering plants was attained. He cites several cases of plants which are dioecious, but show indications of a heterostylous ancestry. *Asperula scoparia*, an inhabitant of Tasmania, is dioecious, but the male flowers have large anthers and a very small pistil with rudimentary stigma and style, while the female flowers have a large, well-developed ovary and rudimentary anthers apparently quite destitute of pollen. *Discospermum*, of Ceylon, is apparently heterostyled, but one of the two forms is always barren, the ovary containing about two aborted ovules in each loculus, while in the other form each loculus contains several perfect ovules. The species is therefore really dioecious. Most of the species of the South American genus *Aegiphila* are heterostyled. In *Aegiphila obdurata*, however, the anthers of the long-styled form are entirely destitute of pollen, while the pistil is perfectly developed; in the short-styled form, on the other hand, the pistil is aborted, while the stamens are perfect.

There are a number of facts which indicate (BLAKESLEE 3, p. 371) that in all dioecious plants one sex is dominant and makes its appearance while the other remains latent. Male and female willow plants are frequently found with flowers of the opposite sex. *Lychnis dioica* is normally dioecious, but STRASBURGER (35, p. 692) found in his cultures at Bonn occasional hermaphrodite plants. These were in every case affected by a smut, *Ustilago violacea*, and he attributes the hermaphrodite condition to the action of the fungus. *Ustilago violacea* fruits only in the anthers of the host plant. If it attacks a male plant it fruits in the anthers, and if it attacks a female plant, in some way it stimulates its host to the production of stamens, in which it fruits.

Recently SHULL (32, p. 112) has described occasional hermaphrodite plants occurring in a pure bred normal race of *Lychnis*,

in which *Ustilago violacea* has never appeared.   He reverses (p. 119) STRASBURGER's interpretation of the origin of the diseased hermaphrodites, and suggests that the infected plants were males in which the disease allowed the pistils to develop.   SHULL's discovery that hermaphrodite plants arise occasionally in normal races and his criticism of STRASBURGER's interpretations do not alter the importance of the fact that in a normally dioecious plant the bisexual condition may sometimes occur, perhaps because of some pathological stimulation.   Another instance of the same condition is cited by STRASBURGER (**38,** p. 471).   He reports, in the normally dioecious *Mercurialis annua*, male plants bearing a few female flowers, some of which when pollinated produced good seed.

The condition just described would seem to indicate that dioecious plants arose from the hermaphrodite condition.   If such is the case, dimorphic heterostylous plants, since they already exist in two classes, which differ considerably and are adapted for reciprocal fertilization, might be more likely to become dioecious than would homostylous plants.

In this connection it is tempting to extend LILLIE's view of the origin of sex to the origin of dioeciousness in the higher plants.   He assumes (**23,** p. 375) that fertilization may be always selective, even when there is no morphological gametic differentiation. According to his idea, gametes may be physiologically different even when they are morphologically alike.   Morphological differentiation would then follow naturally, as the expression of these physiological differences, and sex differentiation as a further stage in the same process of evolution.

Is it not entirely probable that different "strains" may exist in some species of hermaphrodite plants which differ in their relations of fertility somewhat as do the different "forms" of heterostylous plants?   Panmixia has always been assumed to be the natural condition of hermaphrodite species.   That is, it has been assumed that any individual can fertilize or be fertilized by any other individual in the species with equal ease, but that such is actually the case has never been proven.   The existence of different "strains" having such relations as suggested above would not be easily demonstrated under natural conditions, as each stigma

doubtless receives pollen from several flowers, among which it could easily "select" the favorable pollen, by inducing a more rapid growth of the pollen tube of the "legitimate" kind, exactly the method by which illegitimate fertilization is prevented, under normal conditions, in heterostylous plants.

The morphological differences shown by the different forms of heterostylous plants are really very slight compared with their physiological differences. According to the view suggested above, dimorphism, trimorphism, and dioeciousness would be merely morphological expressions of physiological differences common to many plants. Such an hypothesis would account for the origin of dioeciousness and heterostyly at different points, widely separated both geographically and genetically throughout the plant kingdom.

Fagopyrum esculentum

The dimorphous flowers of the buckwheat were first described and very accurately figured by Hermann Müller (**28,** p. 165; also **29,** p. 509). As Müller points out, there are "in each form 8 stamens, 3 closely surrounding the styles and opening outwards, the 5 others inserted more outwards and opening inwards." The place occupied in one of the forms by the anthers is occupied in the other by the stigmas, a perfect adaptation for cross pollination by the numerous insects which visit the flowers for the sake of the honey secreted by the 8 globular nectaries at the base of the filaments. There is apparently no difference in the structure of the stigmas in the two forms, but the pollen grains of the short-styled form are larger than those of the long-styled form, their diameters being in about the ratio 5:4. There appear to be no secondary differences in the vegetative structure of the plants.

The flowers are as a rule true to form, and there is no difficulty in distinguishing long-styled from short-styled plants. Occasional flowers with the stigmas at the anther level, however, are seen on normal long-styled plants. Seldom more than one such flower occurs on a plant and this is usually the first which opens. A similar condition has been noted by Bateson and Gregory in *Primula* (**2,** p. 583). One plant, however, was noted among some grown

for experimental purposes in the greenhouse, which showed such unusual floral variations as to be worthy of record. The plant produced 10 blossoms, and 8 of these had stamens and pistil both long; while 2, the third and eighth, were normal short-styled flowers. The earlier blossoms were removed in order to secure continued flowering. Later, however, two of the abnormal flowers were pollinated, one with pollen from a short-styled and the other with pollen from a long-styled flower, but neither developed seed. One of the normal short-styled flowers on this plant, however, produced a good seed when self-pollinated.

It will be noted that in both the cases cited above, in which the stamens and pistil are of the same length, it is the pistil which has varied from its normal length. That is, if a flower on a short-styled plant shows pistils and stamens of equal length, both are long; while in such a flower on a long-styled plant, both pistil and stamens are short. A curious case which shows a similar variation in the length of the pistil has been noted by BATESON and GREGORY (2, p. 583) in *Primula sinensis*. There is a variety with a very large yellow "eye" extending up over the limb of the corolla, quite distinct from the small yellow pentagon characteristic of the normal flower. This variety has the anthers in the position normal for long-styled plants, but the style is short and the stigma just reaches the anther level, a condition which BATESON and GREGORY designate as "equal-styled." In investigating the inheritance of these two unusual characters, they find that the "equal-style" is the form which the long-styled type assumes when the plant is homozygous in the large eye character. In this case, as in the others mentioned, the pistil alone varies from the normal length.

DARWIN experimented in a rather imperfect manner on the relative fertility of the two forms in the buckwheat, and showed that illegitimate fertilization is less successful than legitimate, yielding fewer and smaller seeds. In order to gain some knowledge of the relative ease of legitimate and illegitimate fertilization, the following experiment was made. Plants were grown from seed in a greenhouse, where there were no insects which might bring about pollination. The flowers which were to be experimented upon were examined with a lens in order to make sure that the pistils

had not been accidentally pollinated, and were castrated to pre-
vent self-pollination. They were then pollinated artificially, either
with pollen from a plant of the other form "legitimately," or with
pollen from another plant of the same form "illegitimately."

After a definite number of hours, the pistils were fixed and
microtone sections prepared. In the case of legitimate pollination,
pistils fixed 18 hours after pollination showed regularly a 3-celled
pro-embryo, and at least three free nuclear divisions had occurred
in the endosperm. The embryo was usually in the quadrant stage
24 hours after legitimate pollination. The time elapsing between
pollination and fertilization seems to have no relation to the dis-
tance traveled by the pollen tube, for it did not differ perceptibly
in the two forms.

Pistils which had been illegitimately pollinated showed when
sectioned that in 24 hours the pollen tube had made but a very
slight growth. After 48 hours there was a greater development of
the pollen tube; and in 3 days (72 hours) a few pistils showed the
pollen tube extending nearly to the egg. Some of the pistils which
were fixed 96 hours after illegitimate pollination showed the embryo
in the 8- or 16-celled stage.

Under the conditions of the experiment then, if pollen from either
form was placed on the stigma of a flower of the other form, the
growth of the pollen tube and the fusion of the two nuclei required
considerably less than 18 hours. But if pollen from either form
was placed on the stigma of a flower of the same form, even though
on a different plant, a period longer than 3 days was required for
the tube to reach the egg. Illegitimate fertilization would then
practically never occur in nature, especially in a form so frequently
visited by insects. H. MÜLLER records (**28,** p. 165) 41 species of
insects seen on the flowers of the buckwheat, many of them very
frequently.

It is entirely possible that the rate of growth of the pollen tube
was more rapid under the conditions of the experiment than it is
in nature, for the temperature of the greenhouse in which the plants
were grown was rather high. It does not seem probable, however,
that this would affect the relative rate of growth of the pollen tube
in the two cases.

THE REDUCTION DIVISION OF THE POLLEN MOTHER CELLS

Each loculus contains usually a single row of 8 or 10 pollen mother cells. All the cells of each loculus apparently pass through the different stages simultaneously, and there is no evidence of a regular basipetal succession in their development, such as has been reported in numerous cases. It was necessary, therefore, in order to determine the succession of the various stages, to compare carefully the cells of different loculi, using their size and the condition of their cytoplasm as a check upon the order of the phases shown by the nuclei. Considerable variation in different loculi of the same flower is common.

PROPHASE.—The pollen mother cells first become distinguishable by their increased size and the possession of a large nucleus, containing a single large, dark staining nucleolus (figs. 1, 2). This nucleolus is surrounded by a clear zone which is apparently not affected by any of the stains used. In a few cases two such nucleoli were observed in one nucleus, each surrounded by the colorless area just described. The nature of this clear space seems to be rather uncertain. It has been figured frequently, and is regarded by some writers as a constant structure. MARTINS MANO (24, p. 60) speaks of it as the "peri-nucleolar vacuole." STRASBURGER, however, considers that the appearance is due to reagents (37, p. 519).

Surrounding this clear zone is the nuclear reticulum, consisting of very delicate indefinite threads which do not take the chromatin stain. This "linin" network contains scattered dark staining bodies which are apparently rather irregular in number and do not seem to occur in pairs. They thus furnish no support for the attractive prochromosome theory of ROSENBERG (31, p. 25) and others.

SYNAPSIS.—The recent work of LAWSON (22) has again raised the question as to whether the phase of the nucleus preceding the reduction division, characterized by an apparent condensation and contraction of the chromatin on one side of the nuclear cavity, is a real contraction. For some time this condition was regarded as an artifact, but it has been observed in living material by several investigators, and its occurrence, at least in some forms, is now

generally admitted. Some writers, however, still regard it as due
to imperfect fixation.[1] This stage was first called "synapsis" by
MOORE in 1895; and many cytologists have come to regard it as
an important and critical stage, when the actual fusion of the
maternal and paternal chromosomes occurs.

LAWSON presents a different explanation of the condition
observed. He interprets the phenomenon as simply a growth
period of the nucleus, during which the increased osmotic pressure
within the nucleus causes the absorption of a considerable amount
of cell-sap, and the consequent increase in size of the nucleus. In
this enlargement the chromatin mass is left behind. The char-
acteristic position of the chromatin mass at one side of the nucleus,
according to LAWSON, is due to the fact that the extension of the
nuclear cavity always takes place in one direction, that is, on the
side toward an intercellular space where there is least resistance
from the neighboring cells.

Since the publication of LAWSON'S paper, the writer has studied
the synaptic stages in the buckwheat with special reference to the
comparative size of the nucleus and chromatin mass before and
during synapsis. There is certainly an increase in the size of the
nucleus during the synaptic stages, as will appear from a compari-
son of figs. 3, 4, and 5. It seems equally certain that the chromatin
mass occupies a much smaller space during the "balled-up" con-
dition than it does either before or after this stage. The stage is
evidently of considerable duration, longer than all the later stages
in the heterotypic division combined.

It is of course possible that the contraction is due to imperfect
fixation, that the nuclear matter is for some time in such a condition
that it is impossible to preserve its structure by any known method.
A contraction at this stage, however, is of constant occurrence, and
the chromatic material has a characteristic appearance after the
"contraction" which differs markedly from its appearance before.
The same conditions were found in *Houstonia;* and in the following
descriptions it will be assumed that synapsis is a normal stage.

A comparison of the nuclei before and after synapsis is rendered

[1] A full discussion of this subject, together with citations of literature, is given by
GRÉGOIRE (**17,** pp. 332–335).

easier by a change which takes place in the cell at this time, and
by which postsynaptic stages are clearly distinguished from pre-
synaptic stages.   During the contracted condition of the chromatic
mass the pollen mother cell becomes rounded and takes on a more
spherical form.   In figs. 3, 4, and 5, the nuclei are apparently in
much the same stage, but the cells show a progressive "rounding
off."   Such a change in the shape of the cell during synapsis has
been observed by STRASBURGER, ALLEN (1, fig. 19), DAVIS (12,
p. 634), and others, and appears in the figures of many workers
who make no particular mention of it.

The synaptic contraction seems to take place by a drawing
together, at one side of the nucleus, of the whole nuclear reticulum,
usually but not always including the nucleolus (figs. 3 and 5).   On
careful examination this mass appears to consist, at least in part,
of delicate threads; but no evidence of any pairing of these threads,
such as has been described by some investigators, could be obtained.
The threads of the nuclear reticulum in the buckwheat are so
delicate, however, that it would be extremely difficult to demon-
strate any such condition even if it occurred; and the behavior
of the chromatin at later stages makes it seem probable that a pair-
ing has actually taken place at this stage.

The amount of chromatin staining material is very markedly
increased during synapsis.   Before the contraction, the greater
part of the reticulum does not take the chromatin stain; but the
nuclear mass comes out of the contracted condition as a series of
rather thin loops (fig. 7), each of which consists, apparently, of a
single thread which takes the chromatin stain uniformly throughout
its length.   This thread is granular in appearance and varies some-
what in thickness; but no alternation of chromatic and achromatic
material, such as has been described by some writers at this stage,
could be made out.   The number of loops is rather inconstant,
but is generally greater than the number of gemini.

These loops gradually shorten and thicken (fig. 8) and become
more dense and uniform in appearance.   At the time of greatest
thickness they undergo a longitudinal split (fig. 9), thus giving rise
to a series of paired chromatic threads from which the gemini
are apparently formed by the continued gathering together of the

chromatic material. The stages from the loosening of the synaptic knot to the formation of the gemini are passed through rather rapidly; and the changes apparently take place in all parts of the nucleus at practically the same time. There does not seem to be any definite "second contraction," but rather a continuous shortening and thickening of the threads from the time the thin loops first appear until the gemini are formed.

The two members of a geminus are generally united at one end, but they are often found entirely separate or united throughout their length (figs. 11 and 12). Their appearance in figs. 10 and 11 would indicate that the loops from which they arose consisted of both chromatin and linin, and that in the formation of gemini at least a part of the linin is discarded. Diakinesis is apparently of considerable duration and affords an excellent opportunity for counting the chromosomes. The reduced number is evidently 8. The nucleolus appears at this stage as a pale and somewhat irregular body (figs. 10–12).

The succeeding stages present no unusual features. The first division separates the two members of the gemini, but there is no evidence of a longitudinal split in the chromosomes during the anaphase (fig. 13). During interkinesis the daughter nuclei of the first division approximate somewhat a resting condition. A rather definite nuclear membrane is formed, a pale nucleolus appears, the chromosomes become more or less vacuolate, and are connected to some extent by indefinite linin threads (fig. 18). It is usually possible at this stage, however, to make out the separate chromosomes and to determine definitely the reduced number, 8. The second division is a typical homotypic division.

It will be noted that the condition described for the buckwheat corresponds closely with the "hétérohoméotypique scheme" of GRÉGOIRE (**17,** p. 233). His "scheme" may be briefly outlined as follows: In the early prophase (p. 243) of the reduction division, the nuclear mass becomes resolved into a number of fine threads, each of which is the equivalent of a somatic chromosome. This is the leptotène stage. These threads, the "gamomites," become arranged in pairs (zygotène stage), which afterward fuse to form a series of independent loops, the pachytène loops. These pachy-

tène loops exist in the haploid number, and soon undergo a longi-
tudinal division, which is really the separation of the parts which
united in their formation. The halves thus separated often appear
irregularly spread apart and crossed, the "strepsitène" stage;
and the "gemini" are formed by the shortening and thickening
of these strepsitène loops. Diakinesis (p. 232) is characterized
by the presence of "chromosomes" in the reduced or haploid
number, formed often of two rather independent branches. The
first division separates the two branches of these "chromosomes,"
and the daughter chromosomes show during the anaphase of the
first division a longitudinal split, which is sometimes visible in the
two branches of the diakinetic "chromosomes." After an inter-
kinesis, more or less brief, marked by varying degrees of nuclear
reconstruction, the daughter chromosomes of the first division
reappear, and their longitudinal halves are separated in the second
division.

In GRÉGOIRE's scheme, synapsis, when it occurs, is due to the
contraction which accompanies the fusion, in pairs, of the lepto-
tène threads to form the pachytène loops. If this change takes
place simultaneously throughout the nucleus, a crowding of the
whole chromatin mass at one side is the result.

If it is assumed, as seems probable, that in the buckwheat a
fusion of thin filaments takes place in the presynaptic stages, then
the series of loops characteristic of the early postsynaptic period
represents a pachytène stage. There are, however, considerably
more than the haploid number of loops. This may mean either
that the loops representing the chromosomes are long (GRÉGOIRE,
p. 335) or that they are parts of a continuous spirem thread. These
two explanations represent the two interpretations of the spirem
condition, STRASBURGER and his school maintaining that the
chromatic mass comes out of synapsis as a continuous spirem thread,
GRÉGOIRE holding that the so-called spirem is really a series of
independent loops. On this point the buckwheat furnishes no
evidence. It seems certain, however, that these loops become
shortened and thickened and undergo a longitudinal split, forming
the strepsitène loops, from which the gemini are derived by a
continued thickening.

It was also noted above that no split in the chromosomes is evident in the anaphase of the first division. But though such a split is stated by GRÉGOIRE to be a part of his "scheme," its absence cannot be regarded as a very important deviation.

THE CHROMOSOMES IN THE REDUCTION DIVISION

As the chromosomes become vacuolate during interkinesis, the stage most favorable for an examination and comparison of the chromosomes is the anaphase of the reduction division (fig. 13). The most striking thing about the chromosomes at this stage is their different size in the two forms. The chromosomes of the short-styled form have a diameter nearly twice as great as do those of the long-styled form (compare figs. 14 and 15 with figs. 16 and 17). As it is entirely improbable that this difference in size can be due to any difference in the hereditary qualities borne by the two sets of chromosomes, it appears to be related with the corresponding, though smaller, difference in the size of the microspore mother cells at this stage.

Just what relation exists between the size of the cell, the size of the nucleus, and the size and mass of the chromosomes is not well understood. There seems, at any rate, to be no definite relation between the number of chromosomes and the size of the nucleus. STRASBURGER (**36,** p. 51) cites several cases in which the nuclei with the diploid number of chromosomes are distinctly larger than those with the haploid number in the same species. In the seed rudiments of *Taxus baccata*, for example, the nuclei of the pro-thallium are much smaller than those of the nucellus. That the diploid number of chromosomes is not always associated with a larger nucleus than that which contains the haploid number, how-ever, he proves by the case of *Dictyota dichotoma*, in which species the nuclei of the plants which produce tetraspores are no larger than those of the plants which bear eggs or sperms, though they have, of course, twice as many chromosomes.

Another difference between the two forms, which is apparently constant, is the arrangement of the chromosomes in the anaphase of the heterotypic division. In the short-styled form the eight chromosomes tend to be arranged with six in the peripheral ring

and two in the middle; while in the long form the arrangement shows seven in the periphery and one in the middle. This arrangement may of course be accidental or it may be mechanical, due to the difference in the chromosomes (compare figs. 14 and 15 with figs. 16 and 17).

A careful examination of the cells in the late anaphase of the heterotypic division of the short-styled form failed to show any difference in the chromosomes (figs. 14 and 15). In the long-styled form, however, the "central" chromosome is apparently considerably larger in one of the daughter nuclei of the heterotypic mitosis than is its synaptic mate in the sister nucleus (figs. 16 and 17). While this condition is apparently constant, little importance can be attached to it until more is known of the inheritance of heterostyly in the buckwheat,[2] and of the reduction division of the megaspores. It bears a striking resemblance, however, to the condition found in the sperm mother cells of *Lygaeus* and other insects (WILSON, **40,** p. 59) in which there is an "x" chromosome which has as a synaptic mate a smaller "y" chromosome.

GROWTH OF THE POLLEN MOTHER CELLS

The difference in the size of the pollen grains of the two forms has been referred to above. As a corresponding difference in size was apparent in the pollen mother cells during early stages, an examination was made to determine whether a similar difference in size occurs in the somatic cells. Buckwheat seeds were allowed to germinate on moist filter paper, and when the roots were about half an inch in length the tips were removed with a sharp razor and fixed at once. Seeds and root tips were carefully marked to correspond and the seeds planted.

The seedlings developed readily, and when the plants blossomed, microtome sections of the root tips were prepared and the size of the embryonic cells of the two forms compared. The cells measured, of course, were always in the same stage, usually the metaphase. Some variation in the size of the embryonic cells even in the same stage was noted, but no constant difference between the two forms

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[2] DARWIN's experiments on this species, as he himself states, were very imperfect, and gave the result that illegitimate seeds of either form produced plants of both forms.

could be demonstrated. No difference is apparent either in the rest-
ing nuclei of the pollen mother cells or in the pollen mother cells
themselves (figs. 1 and 2). The difference becomes evident, however,
after the rounding off of the pollen mother cells during synapsis,
and apparently reaches its maximum some time before diakinesis,
(compare figs. 7 and 8; and figs. 11 and 12). It is of course
much easier to measure accurately spherical cells than angular cells
massed together, as it is extremely difficult to determine in the
latter case whether the cells are cut in the same plane or not, and
there may be a difference in the size of the somatic cells which could
not be determined. It seems entirely probable, however, that the
difference in the size of the pollen of the two forms is due to the
fact that the pollen mother cells of the short-styled form grow
more rapidly during the period from the beginning of synapsis to
diakinesis, than do the microspore mother cells of the long-styled
form. It is interesting to note in this connection that it is during
this same period that the ovocyte in animals undergoes its great-
est enlargement (GRÉGOIRE, **17,** p. 243).

SEPARATION OF THE POLLEN GRAINS

The separation of the pollen grains occurs in much the same
way as was described by Miss FERGUSON for *Pinus* (**14,** p. 35).
During the late telophase of the second mitosis in the microspore
mother cell the four nuclei of the tetrad become connected with
one another in all directions by "kinoplasmic" fibers (fig. 19).
These fibers, however, are never very numerous, and they are visible
for only a comparatively short time after the reconstruction of the
daughter nuclei is complete. During this period a marked thick-
ening of the wall of the pollen mother cell occurs (fig. 20); and,
apparently continuous with this wall and extending out from it,
walls appear separating the daughter cells (fig. 21).

These walls attain a remarkable thickness, and are apparently
homogeneous and extremely resistant to stains. They stain very
lightly with haematoxylin or safranin, and with orange G only if
the staining be considerably prolonged. The strength and definite-
ness of the walls thus formed is shown by the fact that after the
spores are mature and the wall of the mother cell is ruptured, the

empty mother cell wall with its four chambers often persists for some time (fig. 22).

TAPETAL CELLS

About the time the pollen mother cells reach the pachytène stage, the tapetal cells begin a rather irregular free nuclear division. The nuclei show all gradations from true mitosis to what is apparently simple amitotic division. Figs. 23–25 show the amitotic division of these nuclei. By the time the pollen mother cells have finished the homotypic division, the tapetal cells regularly contain two and sometimes four free nuclei (fig. 26).

MEGASPORE MOTHER CELLS

The difficulty of orienting the buds makes the number of preparations necessary to secure a full series of stages so great that a complete study of the development of the megaspores has not yet been made. One preparation of the long-styled form, however, showed typical diakinesis, which, as was to be expected, had much the same appearance as that in the microspore (fig. 27). Usually only one megaspore mother cell is formed in an ovule, and each flower produces but a single seed. One ovule, however, which happened to be in the long-styled form, contained two apparently well-developed and normal megaspore mother cells, with their nuclei in the pachytène stage. Their shape seems to indicate that they were formed by the division of a single cell by an anticlinal wall (fig. 28).

Houstonia caerulea

The Rubiaceae contain nearly half the genera known to be heterostylous. The flowers of *Houstonia* are plainly dimorphic, the pistil being exserted in one form and the stamens in the other. The pollen grains vary somewhat in size in each form, but those of the short-styled form are larger than those of the long-styled, their diameters being in about the ratio 10:7. No differences have been noted in the vegetative structures of the two forms. The flowers are so small that experimenting with them would be very difficult, and accordingly very little is known about the relative fertility of legitimate and illegitimate unions. DARWIN, however (**11,** p. 132),

observed that some short-styled plants growing by themselves at a considerable distance from any long-styled plants produced mostly sterile capsules. From this he concludes that the short-styled form is very sterile with it own pollen.

The pollen mother cells are small and a large number are contained in one loculus, often as many as 40 appearing in a single longitudinal section. Considerable variation is generally shown by the cells of a loculus, but there does not seem to be any very regular succession of stages. Frequently, to be sure, an anther shows a progressive series with the most advanced stages at the top; but this is by no means a uniform condition, for occasionally a loculus shows the mother cells near the middle in a more advanced condition than the cells at either end.   The relative position of the cells in an anther does not, then, in *Houstonia*, furnish reliable evidence of the succession of the stages.   This makes the exact significance of some stages rather uncertain, and some of them are open to more than one interpretation.

THE REDUCTION DIVISION OF THE POLLEN MOTHER CELLS

PROPHASE.—The pollen mother cells first become distinguishable by their increased size and the possession of a very large nucleus. The nuclear reticulum appears as a network of very fine irregular threads, and contains numerous granules, none of which take the chromatin stain.   There is usually only a single large nucleolus, and this is surrounded by the clear zone already described in the buckwheat.

The nuclei of the pollen mother cells present at this stage an appearance which has been variously interpreted.   Figs. 29–33 show what appears like a progressive "budding off" of chromatin staining material from the nucleolus.   A similar condition has been observed by DARLING (**5,** p. 184) in *Acer Negundo*, and described as a budding off of actual chromatin which goes to make up the spirem thread.   Miss NICHOLS (**30,** p. 35) has observed this condition in *Sarracenia*, and considers that it represents a movement of chromatin, which has been elaborated in the nucleolus, to the nuclear reticulum.   GATES (**15,** p. 6) interprets a similar appearance in *Oenothera* in an entirely different manner.   He regards the

dark staining bodies in the nuclear reticulum as small nucleoli, and thinks that conditions similar to those in figs. 30, 32, and 33 represent a fusion of some of these smaller nucleoli with the large one.

As it is impossible to arrange these stages in *Houstonia* with any certainty, there is no proof as to what actually takes place. The appearance strongly suggests a "budding off" of material which is caught up by the nuclear reticulum; and the presence of numerous dark staining bodies in the reticulum at the time of synapsis (figs. 34 and 35) have been held to show that such is actually the case. There is no proof, however, that the dark staining bodies which are present in the reticulum at this time have any connection with the nucleolus. In fact, a series of somatic stages, taken from the rapidly growing tissue of a young ovule (figs. 36–42), seems to show that the dark staining masses appearing in the reticulum previous to the formation of the spindle have no connection, at least directly, with the spherical bodies observed near the nucleolus, but that they are chromosomes which become differentiated from the nuclear reticulum during the prophase.

SYNAPSIS.—Synapsis is characterized by a crowding together at one side of the nucleus of the entire nuclear reticulum (figs. 34 and 35). No structure can be made out in this mass except that it consists of a number of dark staining bodies in a much lighter, rather indefinite network. Synapsis is of considerable duration, and during this period the nucleus undergoes a marked increase in size (compare figs. 34 and 35) and the cell becomes rounded (fig. 43).

SPIREM STAGE.—The chromatic mass comes out of synapsis in a series of thin loops (figs. 43 and 44). Each loop apparently consists of a single thin thread which does not take the chromatic stains uniformly throughout its length, but shows numerous dark staining bodies, connected by paler linin portions. As these loops shorten and thicken they stain more uniformly (fig. 45).

The synaptic knot loosens very slowly, and even at the period of greatest thickening of the loops there is still a considerable portion of the nuclear mass, which does not show any definite structure, surrounding the nucleolus (fig. 45). This condition can be explained only on the supposition that some parts of the nucleus often pass

through the various stages in advance of others, a condition comparable to that noted by JANSSENS (**21**) in *Batracoseps*. The appearance at later stages seems to bear out this supposition. Fig. 46 shows a split in a portion of the spirem, while the rest appears entirely undivided. Fig. 47 shows several places where the parts have become still more widely separated, yet a considerable mass of the nuclear material is still in synapsis; and fig. 49 shows all gradations from paired loops to typical gemini.

This irregularity in development, together with the fact that there is no definite succession of stages in the loculi, makes a fully satisfactory interpretation of the spirem stages impossible. If it is true, however, as seems probable, that the chromatic loops which appear as the mass emerges from synapsis consist of a single thread[3] which afterward shortens and thickens, and if the split shown in figs. 46 and 47 represents a separation of threads previously paired, the series accords closely with GRÉGOIRE's hétérohoméotypique scheme. Figs. 44 and 45 would then represent the pachytène stage, the loops shown in fig. 45 resulting from a shortening and thickening of thinner loops shown in fig. 44. Fig. 46 doubtless represents a splitting of the pachytène loops, the diplotène stage. Figs. 47 and 48 show different stages in strepsinema; in fig. 47 only a small part of the nuclear mass is in the strepsitène condition, while fig. 48 is a more advanced stage.

The appearance of fig. 43, which is quite characteristic of the loosening of the synaptic knot, makes it appear possible that the spirem comes out of synapsis as a series of paired threads which afterward fuse to form a continuous spirem. That this actually occurs in most cases is held by STRASBURGER and his school. Such a condition as is shown in fig. 44, however, where the loops appear still thin but with the halves widely separated, makes it seem probable that the thickened spirem arises from the thinner by a thickening of the threads. That is, figs. 44 and 45 represent merely different phases of the pachytène stage.

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[3] In referring to this stage as characterized by the occurrence of loops consisting of a single thread, the writer does not mean to enter into the discussion as to whether this "single thread" is really a unit or is composed of two separate threads twisted together. The expression "single thread" is used to mean simply a loop which appears as one thread, as distinguished from one composed of two parallel threads.

DIAKINESIS.—In either case the diakinetic gemini arise by the continued shortening of the paired loops which make up the strepsitène stage. As shown by fig. 49, diakinesis may not arise simultaneously throughout the nucleus. The stage is of considerable duration, however, and presents a very characteristic appearance (figs. 50 and 51). The chromosomes of a geminus are generally united only at one end and often diverge widely from one another. The nucleolus at this stage appears vacuolate, and shows in section an outer dark staining region surrounding an inner almost colorless portion. Commonly at this stage one or two gemini appear clinging to the nucleolus (fig. 51). Their appearance suggests the condition reported by DARLING (5, p. 186) for *Acer Negundo*, where five chromosomes appear to arise directly from the nucleolus. There is nothing in *Houstonia*, however, to indicate that this condition is anything more than a clinging of the gemini to the nucleolus.

Another characteristic appearance during diakinesis is that shown in figs. 52 and 53, where several gemini appear clinging together in a single row. This condition very much resembles that found by GATES (15, p. 12) in *Oenothera*. GATES, however, considers that in *Oenothera* this condition is previous to true diakinesis, and represents a single continuous spirem constricted at regular intervals to form a chain of chromosomes. Some of these chromosomes afterward pair to form typical gemini, but a considerable number of them are apparently taken up by the heterotypic spindle without having previously paired. An essentially similar method of formation of the diakinetic chromosomes has been reported by GEERTS (16, p. 610) and by Davis (12, p. 559) for *Oenothera*, and by YAMANOUCHI (41, p. 186) for *Fucus*.

That such is the case in *Houstonia*, however, seems improbable, as such an interpretation makes it necessary to regard the condition shown in figs. 46 and 47 as a precocious split which afterward closes up. Moreover, the two members of each geminus in *Houstonia* are almost always found attached at one end, while in *Oenothera* such a condition is the exception. The condition shown in figs. 52 and 53 seem to be best explained as a temporary union of independent gemini, an interpretation first suggested by MIYAKE (25, p. 96) for a similar appearance in *Galtonia candicans*.

INTERKINESIS.—The heterotypic division presents no unusual features, but interkinesis differs markedly from that of the buckwheat. Although a rather definite nuclear membrane is formed, the chromosomes show no signs of vacuolation or anastomosis. On the contrary, they become arranged around the periphery of the nucleus and present at this stage the most satisfactory opportunity for counting and comparing the chromosomes (figs. 57 and 58). The haploid number is 16.

Nucleoli appear at this stage and stain with haematoxylin in exactly the same way as the chromosomes themselves; in fact they were at first mistaken for larger, more regular chromosomes. With safranin, however, the nucleoli are clearly differentiated from the chromosomes. There may be either one or two nucleoli at this stage, but sister nuclei seem to agree in this respect, that is, if one daughter nucleus of the first division shows one nucleolus, its sister nucleus also has only one, but both may, on the other hand, have two nucleoli (figs. 57 and 58). This peculiarity seemed at first to bear out the idea that they were chromosomes, and there seems to be a constant difference between the two forms, the long-styled form having two nucleoli (fig. 58) and the short-styled form one larger nucleolus (fig. 57). This may possibly be due to the different sizes of the nuclei in the two forms.

THE CHROMOSOMES IN THE REDUCTION DIVISION

Interkinesis, because the chromosomes remain apparently unchanged and are arranged in the periphery of the nucleus, affords the best opportunity for comparing the chromosomes of the reduction division. No difference in the chromosomes that enter into the formation of the daughter nuclei could be discovered. In fact, both in interkinesis and in the anaphase of the reduction division the chromosomes show very little variation in size. A constant difference in size between the chromosomes of the long-styled and short-styled forms is evident in the anaphase, but this is much less marked than in the buckwheat (compare figs. 55 and 56).

RELATIVE SIZE OF THE POLLEN MOTHER CELLS

The pollen grains of the two forms differ fully as much in size as do the two forms in the buckwheat, but the difference does not

seem to appear so early in their development. The fact that the cells are small and vary somewhat in size makes it difficult to determine when the difference becomes most pronounced; but it will be clear from a comparison of the figures that at diakinesis, the heterotypic division, and the formation of the tetrad, some difference in size is evident. Compare figs. 50 and 51, 55 and 56, and 59 and 60.

<div align="center">SEPARATION OF THE POLLEN GRAINS</div>

The daughter nuclei of the homotypic division show at an early stage a nucleolus similar in staining reactions to that of inter-kinesis (figs. 59 and 60). That is, the nucleoli stain with hema-toxylin exactly the same as the chromosomes, so that in early stages of nuclear formation it is impossible to distinguish them, and the nucleolus appears to arise by a fusion of the chromosomes. Staining with safranin, however, shows clearly that the nucleolus arises separately, but increases in size as the chromosomes lose their staining capacity.

The separation of the pollen grains takes place in much the same way as in the buckwheat, except that in *Houstonia* the "kino-plasmic" fibers connecting the nuclei of the tetrad are much more clearly marked and persistent. The walls which surround the mother cells and separate the cells of the tetrad are not so thick or resistant as in the buckwheat.

Summary of observations

Fagopyrum esculentum

The flowers, as a rule, are true to form, but occasional "equal-styled" flowers are found on both long-styled and short-styled plants. None of these "equal-styled" flowers have been proved to be fertile.

In the case of legitimate pollination less than 18 hours is required for the growth of the pollen tube and the fusion of the egg and sperm nuclei. Illegitimate fertilization is possible, at least in part of the cases; but in case of illegitimate pollination more than 72

hours is necessary for the growth of the pollen tube and the fusion of the nuclei.

No evidence of prochromosomes was found in the pollen mother cells.

The formation of the gemini and the reduction division apparently follow the hétérohoméotypique scheme of GRÉGOIRE; but no split in the chromosomes is evident in the anaphase of the first division.

The reduced chromosome number is 8.

In the anaphase of the reduction division of the microspore mother cells the chromosomes of the short-styled form have a diameter nearly twice as great as do those of the long-styled form.

At this stage the chromosomes of the short-styled form are arranged six in the peripheral ring and two in the middle; while those of the long-styled form are arranged seven in the periphery and one in the middle.

In the long-styled form the "central" chromosome of one of the daughter cells of the first division appears to be larger than its synaptic mate.

Interkinesis is characterized by a partial reconstruction of the nuclei, a nucleolus appears, and the chromosomes become somewhat vacuolate but never lose their identity.

A difference in size of the pollen mother cells of the two forms, corresponding to the difference in the size of the pollen grains, is evident at diakinesis. This difference apparently arises through the greater growth of the pollen mother cells of the short-styled form up to this stage, for no difference in the size of the somatic cells can be found.

At the separation of the cells of the tetrad a thick wall, apparently homogeneous and extremely resistant to stains, is formed surrounding the pollen mother cells and separating the pollen grains.

As the tapetal cells degenerate, they show free nuclear division which is to some extent at least amitotic.

Usually only one megaspore mother cell is formed in an ovule, but one ovule was found which showed two well-developed megaspore mother cells. These had apparently arisen by the longitudinal division of a single cell.

Houstonia caerulea

The nuclei and chromosomes of the pollen mother cells are so small that many of the prophasic phenomena could not be observed with accuracy, but the formation of the gemini and the reduction division seem to follow the "hétérohoméotypique scheme."

Some portions of the nuclei go through the postsynaptic changes in advance of others.

Diakinesis is characterized by a portion of the gemini arranging themselves end to end in a sort of chain.

The reduced chromosome number is 16.

During interkinesis the chromosomes apparently remain unchanged, and are arranged about the periphery of the nucleus. The nucleoli which appear at this stage resemble the chromosomes when stained with haematoxylin.

No difference is apparent in the chromosomes which separate in the heterotypic division.

In the anaphase of the reduction division the chromosomes of the short-styled form are slightly larger than those of the long-styled form.

The separation of the cells of the tetrad occurs in the same way as in the buckwheat.

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EXPLANATION OF PLATES XXI–XXIII

All figures ×1600

PLATE XXI

Microspore mother cells of *Fagopyrum esculentum*

FIG. 1.—Long-styled form: early prophase.

FIG. 2.—Short-styled form: same stage as fig. 1.

Figs. 3–5.—Long-styled form: successive stages in synapsis.

FIG. 6.—Short-styled form: same stage as fig. 5.

FIG. 7.—Short-styled form: early pachytène stage.

FIG. 8.—Long-styled form: later pachytène stage.

FIG. 9.—Long-styled form: strepsitène stage.

FIG. 10.—Long-styled form: gathering together of chromatic substance in the formation of gemini.

FIG. 11.—Short-styled form: diakinesis.

FIG. 12.—Long-styled form: diakinesis.

Figs. 13–15.—Short-styled form: anaphase of the reduction division.

FIGS. 16, 17.—Long-styled form: anaphase of the reduction division.

FIG. 18.—Long-styled form: interkinesis.

PLATE XXII

Fagopyrum esculentum

FIGS. 19–21.—Long-styled form: successive stages in the separation of the pollen grains.

FIG. 22.—Short-styled form: mother cell wall from which the pollen grains have fallen.

FIGS. 23–26.—Long-styled form: tapetal cells showing successive stages in amitotic free nuclear division.

FIG. 27.—Long-styled form: megaspore mother cell; nucleus in diakinesis.

FIG. 28.—Long-styled form: two megaspore mother cells in a single ovule; nuclei in pachytène stage.

Houstonia caerulea

FIGS. 29–33.—Long-styled form: microspore mother cells in early prophase.

FIG. 34.—Long-styled form: beginning of synaptic contraction.

FIG. 35.—Long-styled form: synapsis.

FIGS. 36–42.—Somatic cells, showing successive stages in growth and division.

PLATE XXIII

Microspore mother çells of *Houstonia caerulea*

FIG. 43.—Long-styled form: loosening of the synaptic knot.

FIGS. 44, 45.—Long-styled form: pachytène stage.

FIG. 46.—Long-styled form: tangential view of nucleus in diplotène stage.

FIGS. 47, 48.—Long styled form: strepsitène stage.

FIG. 49.—Long-styled form: gemini in process of formation.

FIG. 50.—Short-styled form: diakinesis.

FIGS. 51–53.—Long-styled form: diakinesis.

FIG. 54.—Short-styled form: anaphase of the heterotypic division.

FIG. 55.—Short-styled form: polar view of chromosomes in anaphase of heterotypic division.
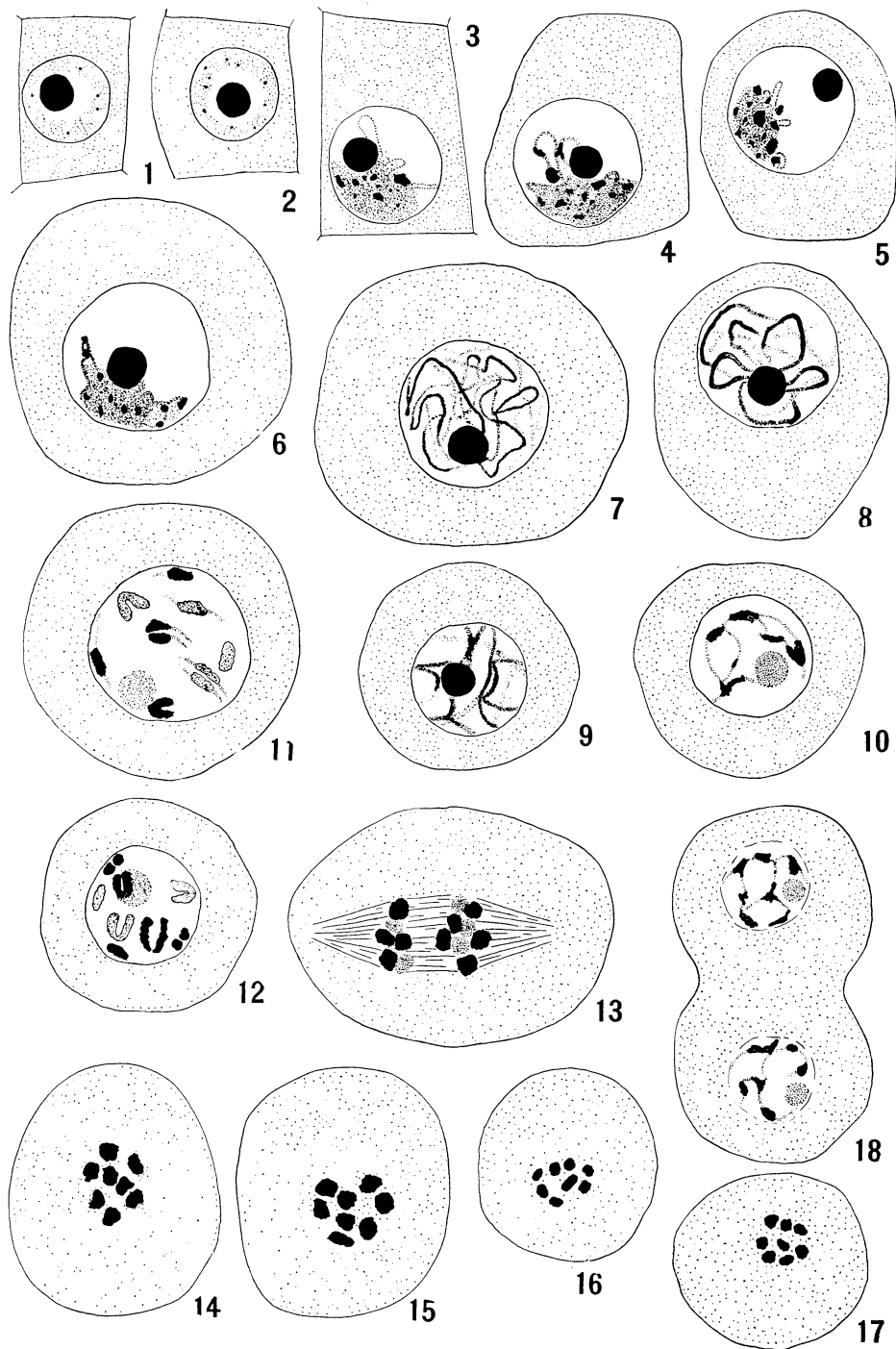
FIG. 56.—Long-styled form: same stage as fig. 55.

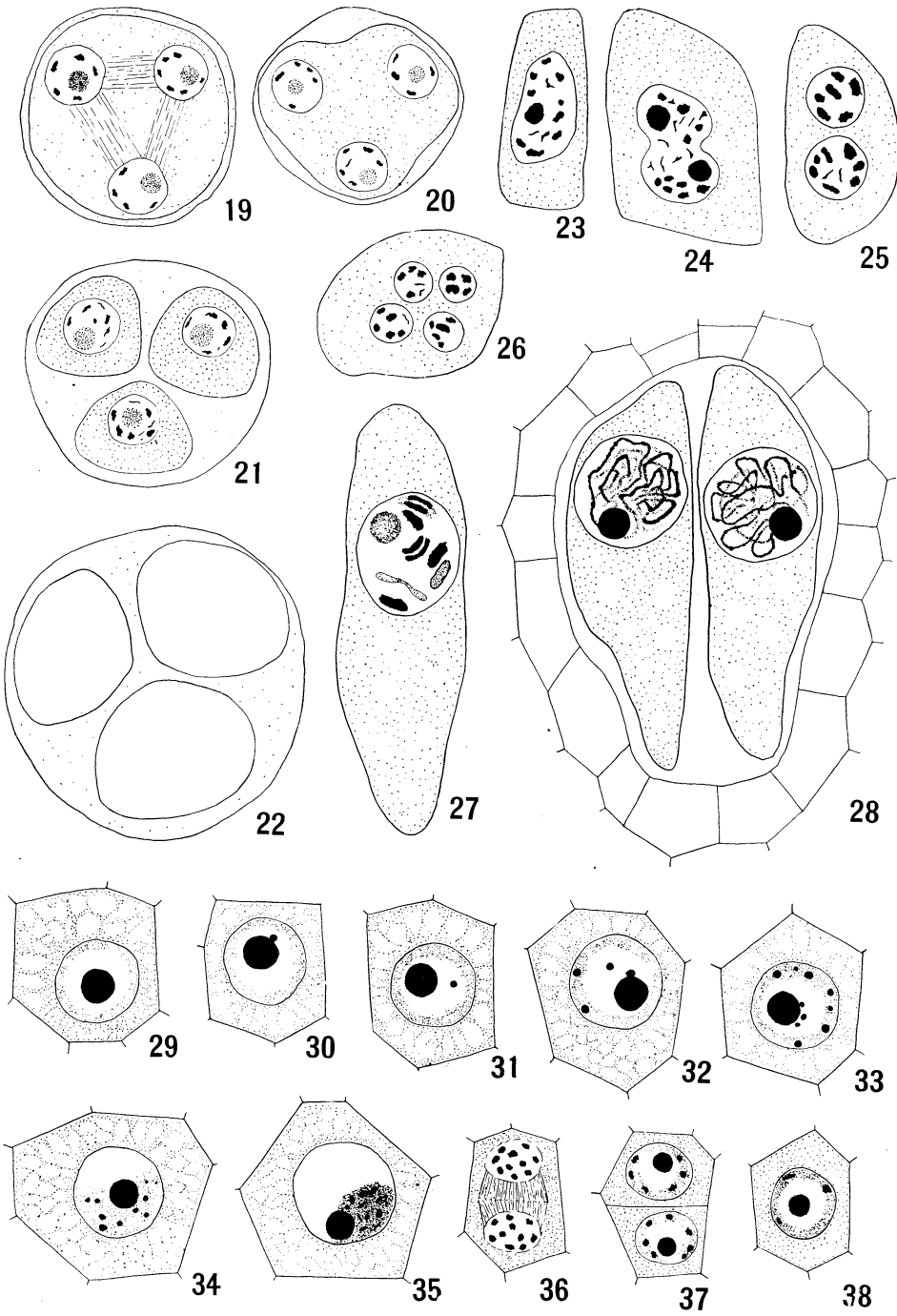FIG. 58.—Long-styled form: interkinesis.

FIG. 57.—Short-styled form: interkinesis.

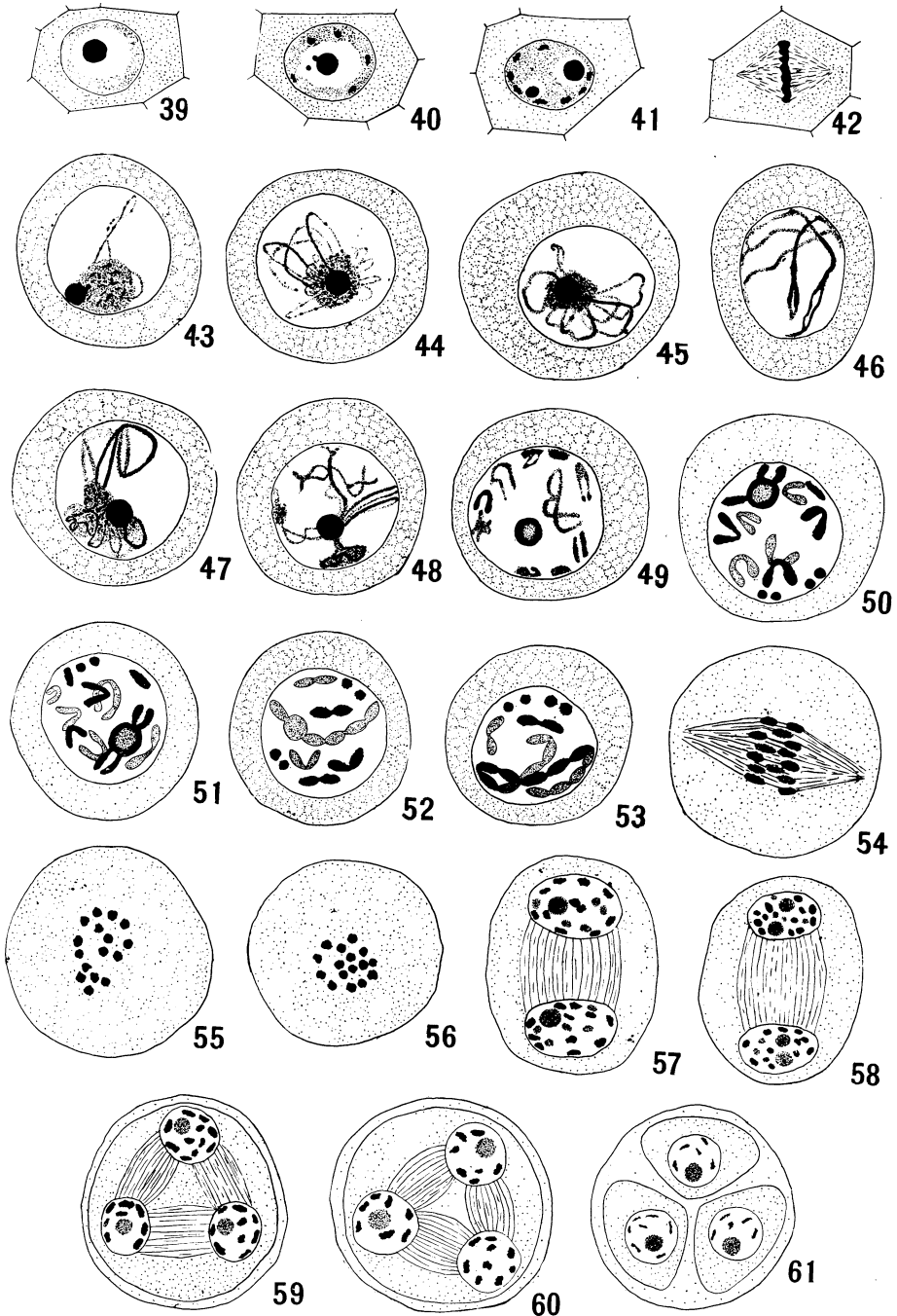FIG. 59.—Long-styled form: reconstruction of daughter nuclei of the homotypic division.

FIG. 60.—Short-styled form: same as fig. 59.

FIG. 61.—Long-styled form: separation of pollen grains.

PLATE XXII

STEVENS on HETEROSTYLOUS PLANTS

PLATE XXIII

STEVENS on HETEROSTYLOUS PLANTS